THE EFFECTS OF TEMPERATURE ON THE DECOMPOSITION OF ALLOCOTHONOUS LEAF LITTER BY GAMMARUS SPP.

ABSTRACT

Global increases in temperature have been shown to have a variety of effects on ecosystems and the organisms that inhabit them on a worldwide scale. Models used to predict future global temperatures show that this climate change is set to worsen, thus increasing the stresses on the natural environment. Through the use of manipulated laboratory microcosms, this study aimed to further examine the effect that temperature increases would have on the ecosystem process of decomposition. It was found that increases in temperature significantly increased the rate of decomposition in aquatic ecosystems, and that this rate of change was consistent across all aquatic ecosystems. Salinity inhibited an increase in the rate of decomposition at lower temperatures, but at higher temperatures the increased rate was comparable to other ecosystems. As global temperatures rise, the functional diversity of detritivores will alter, affecting not only the process of decomposition, but also the amount of energy available in the trophic web. Despite this decomposition rates will increase with temperature, releasing more carbon dioxide and methane into the environment and creating a positive feedback loop for climate change.

INTRODUCTION

It is generally accepted that global temperatures have increased dramatically in the last 150 years, and mean global temperature data from NASA meteorological stations (Figure 1) indicates that the degree of change has increased dramatically in the last 40 years alone. If this observed trend continues, then not only temperatures, but also the rate at which the temperatures change are expected to rise beyond levels experienced in anthropogenic history. Figure 2 displays the degree of change in global surface temperatures over a 46 year period, between 1953 and 1997, (Hansen et al., 1999). From this figure it can be seen that the global areas with the highest degree of change are those with the highest degree of anthropomorphic activity. The cause of this global increase in temperature is a cause of much debate, although many now believe the effects of global change are synonymous with anthropogenic industrial and technological advances, (Begon, Harper and Townsend, 2006).
As temperatures rise the results will be seen on a worldwide scale with, effects being seen on many levels of organisation, from individual organisms through to entire ecosystems. Many biogeographical processes and ecosystem properties are highly temperature sensitive, and the predicted effects of increasing global temperatures include the movement of species ranges - often to cooler, northern areas and the loss of those species unable to migrate or adapt (Chen et al., 2011); a shift towards smaller organism size in ecosystem food webs (Yvon-DuRocher et al., 2010); oceanic acidification and subsequent erosion of calcium carbonate exoskeletons (Anthony et al., 2008); the gradual disassociation of ‘paired’ species phenologies, such as plants and their specialist pollinators (Bartomeus et al., 2011); changes in the composition of communities, such as in those species for which sex determination is temperature dependent (Walther et al., 2002); and the alteration of ecosystem processes and functioning (Mooney et al., 2009).

Metabolism - the process in which organic nutrients such as carbon are consumed and converted to body mass or used in biological processes - is thought to be temperature dependent, and the rate of metabolism increase with temperature is consistent across all organism groups (Gillooly, 2001). As organism metabolism increases, food intake and respiration also increase and thus the carbon dioxide uptake of the whole food web is increased (O’Connor et al., 2009). Atmospheric CO2 levels are known to increase global temperatures, and as well as being released as a by-product of respiration and carbon feedback cycles, increased carbon release from anthropogenic factors such as industry, deforestation and fossil fuel usage, and decreased carbon uptake by the oceans are contributing to significant changes in climate (Wrona et al., 2006; Cox et al., 2000; Sarmiento & Quere, 1996; Schneider & Chen 1980). In addition to increased carbon dioxide, an overall increased metabolism and respiration across all organisms also leads to an increase in other greenhouse gases, such as methane released by methanogenic bacteria during anaerobic respiration (Yvon-DuRocher et al., 2011). Methane has a higher radiative power than carbon dioxide, absorbing more infra-red radiation, and thus being a greater contributor to climate change than carbon dioxide, despite being present in lower proportions (Lashof et al., 1990). The production of methane is more temperature sensitive than the production of carbon dioxide, and so while the proportion of atmospheric methane is currently lower than atmospheric carbon, if global temperatures continue to increase at their current rate then a positive feedback is expected to occur; wherein greater quantities of methane are produced with warming, which in turn will contribute to a global temperature rise (Christensen et al., 2003; Duc et al., 2010; Yvon-DuRocher et al., 2010).

The breakdown of organic matter in an ecosystem by heterotrophic organisms is known as decomposition, and is one of the numerous ecosystem processes that will be affected by climate change as the metabolism of decomposers and detritivores increases in line with the rise in mean global temperatures (Kaiser et al., 2005). The process of decomposition is carried out by decomposers and detritivores including bacteria, fungi and invertebrates, and either releases carbon or methane into the soil and atmosphere, or releases it into the water as dissolved gas in aquatic systems. It is suggested that of all the energy fixed by autotrophic

![Figure 3: A general model of energy flow in a stream, from Begon, Harper and Townsend, 2006.](image-url)
organisms in a benthic environment, more is transferred through the trophic levels by detritivores than by grazers (Lochte & Turley 1988; Silver et al., 1986; Anderson 1979).

Much of the material broken down in an aquatic ecosystem such as a stream or estuary is allochthonous organic carbon, such as terrestrial vascular plant matter which has been deposited in the water from the riparian zone. This organic matter is often washed downstream where it is deposited in piles, or “debris dams”. Here, microbial detritivores begin the decomposition process, impregnating the leaves with inorganic nutrients or accumulating the available nutrients into their own biomass, as well as releasing carbon dioxide or methane into the environment as a result of their respiration (Anderson 1979). The action of the microbial detritivores ‘processes’ the debris for other macro-detritivores such as invertebrates, improving the quality of the leaves as a food source (Romaní et al., 2006). This improved nutritional state is maintained all year round, despite the otherwise seasonal influx of leaf litter into the environment, as different leaf species decompose at different rates, and plant matter is processed at different rates depending on the physical factors which have affected it. For example, abrasions, leaching and microbial colonisation all help to reduce the processing time, (Short & Maslin 1977).

Invertebrate detritivores are known to adopt a range of feeding techniques. For example, filter feeders such as some caddisfly larvae, (Hydropsychidae), filter small particulate debris from the water using nets, while macro-invertebrates such as Gammarus spp. are members of the functional feeding group known as shredders (Brusca & Brusca 2003). They tear parts of the leaf into smaller, more manageable pieces. This allows them to feed, and also releases smaller particles of the leaf into the surrounding water in order that larger filter feeders might also feed, (Kelly et al., 2002). Gammarus spp. are a widespread amphipod crustacean taxa, that are found in all aquatic environments - freshwater, brackish and marine. They occupy the same ecological niche in each system, often proving to be the most dominant benthic invertebrate in that food web, (MacNeil et al., 2007). All Gammarus spp. are laterally flattened, and live in the benthos of the aquatic system. Their morphology allows them to easily move between stones or gravel on the substrate, helping them to avoid being washed downstream by the current. Gammarus pulex is a freshwater species, found in flowing streams with a relatively low organic matter content. They are less tolerant to environmental changes than their brackish counterpart, and so are sensitive to dissolved oxygen levels, salinity, temperature, and a high organic matter content. In contrast, Gammarus zaddachi is found in estuarine waters and intertidal areas. As it is naturally occurring in brackish waters, it is more tolerant to sudden salinity changes and temperature.

From the currently available literature, it seems that increased temperatures have a significant effect on the rate of decomposition across all environments, terrestrial and aquatic, and the most commonly observed trend is that decomposition rates increase with an increase in temperature. However, some studies have observed a decrease in decomposition rate, and this study aims to further investigate the relationship between temperature and decomposition rate across three types of aquatic ecosystem.

We will test the hypotheses that an increase in the environmental temperature will result in an increase in the rate of decomposition - and that the observed trend as a result of this temperature change will be observed equally across all ecosystems - by manipulating the environmental temperature and salinity of laboratory-based microcosms.
METHODS

In this study, three ecosystem types were crossed with three levels of temperature to give rise to 9 combinations of ecosystem type and temperature. Each ecosystem combination was replicated 10 times.

Prior to setting up the experiment, specimens of two species of *Gammarus* amphipod were collected: *Gammarus pulex* from a freshwater river south of the Thames, and *Gammarus zaddachi* from near the mouth of the River Thames, in Deptford. The collected *G. zaddachi* were then divided into two groups, and one group was subjected to gradually increasing salinity levels over a period of several days, in order to prevent osmotic shock once they were placed in water with a higher salinity level. Pre-collected Alder leaves (*Alnus* spp.) were slowly oven dried to remove all moisture prior to the experiment.

Ninety small plastic aquaria were collected, each measuring 0.17 x 0.11 x 0.11 meters, and each was labelled with the designated ecosystem type, corresponding temperature level, and identification number. The pre-collected dried Alder leaves were then weighed using digital scales, and 2.00g (± 0.005g) of leaves were placed into each empty tank. The exact weight of the added leaves was also written on the tank. One litre of the appropriate salinity water was added to each tank - 0 ‰ salinity for the freshwater tanks, 15 ‰ salinity for the brackish tanks, and 30 ‰ salinity for the marine tanks.

Once the tanks were set-up, three individuals of either *G. pulex* for freshwater tanks, or *G. zaddachi* for brackish and marine tanks, were added to each aquarium, and the tanks were taken to three temperature control rooms and kept in a block design in order that the desired temperature be maintained without fluctuations. There, the aquaria were fitted with air-stones to provide aeration for the amphipods and prevent stagnation, and an additional 0.5 litres of the appropriate salinity water were added. The three rooms were kept at their respective temperatures of 5, 10 and 15 °C for the duration of the study.

The tanks were left in the temperature controlled rooms on a 12 hour dark-12 hour light cycle for a total of 49 days in order for the amphipods to begin decomposition the provided leaf litter. They were monitored daily, and any mortalities during the first week were replaced. However, any further mortalities beyond that first week were not. Partial water changes were carried out as required.
After seven weeks, the aquaria were removed from the temperature controlled rooms and transported back to the laboratory where any surviving amphipods were counted and recorded, and the water was drained from the aquaria using a fine-mesh sieve. All remaining leaf matter was removed from the tanks using tweezers and placed into individual paper envelopes, which were labelled with the aquaria details. The leaves were then oven dried at 80 °C for 120 hours. Once the alder leaves were dry, they were then weighed using digital scales, and the following calculation was used in order to calculate the mean rate of decomposition across the duration of the study:

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\text{Initial mass of leaf litter} - \frac{\text{final mass of leaf litter}}{49 \text{ days}} = \text{mean rate of decomposition}
\]

The data were recorded in a tabulated form and then processed using the Minitab16 statistical package to determine the significance of the experimental factors on the rate of decomposition.
RESULTS

Figure 8 below displays the raw data collected during the study in a scatter graph form. A gradual increase in the rate of decomposition can be clearly seen across all ecosystem types as the temperature increases. The rate of this increase appears to be consistent across all ecosystem types also, excluding some anomalous data points which are situated far away from the remaining data. The greatest increase in rate across all ecosystems appears to be at 15 °C, and this temperature also had fewer outlying data points.

![Figure 8: A scatter plot of the raw data collected after 49 days displays an apparently consistent increase in the rate of decomposition with temperature across all ecosystem types.](image)

Figure 9: A bar chart displaying the ecosystem means of the calculated rate of decomposition. Error bars display the standard error of the means.

![Figure 9: A bar chart displaying the ecosystem means of the calculated rate of decomposition. Error bars display the standard error of the means.](image)
When the means of the ecosystem data points are calculated and plotted in a bar chart (Figure 9), a general pattern can be seen wherein the rate of decomposition does increase with temperature, across all ecosystem types. However, the marine ecosystem shows a negative decomposition rate at 5 °C and 10 °C.

A general linear analysis of variance model (Table 1) showed that both temperature and ecosystem type each had significant effects on the rate of decomposition, while the combination of temperature and ecosystem had no significant effect. (GLM ANOVA: Temperature: DF=2, P<0.001; Ecosystem type: DF=2, P<0.001; Interaction: DF=4, P=0.703).

The significance of the temperatures and ecosystem type can be seen in the interaction plot in Figure 10. Of each of the ecosystem types, freshwater had the highest decomposition rate across all temperatures, and the temperatures, 15 °C showed the highest rate of decomposition. However, as the lines in the interaction plot do not cross at any point, there is no interaction between the two factors.
DISCUSSION

There was a significant increase in the rate of decomposition across all ecosystem types and temperatures, but no interaction between the two factors was observed. This suggests that as temperatures rise as predicted by current climate models, then the rate in decomposition will increase and that with no interactions, this increase will be consistent across all aquatic ecosystems. An increase in decomposition rate occurs as a result of an increase in the metabolism of the biota which perform the decomposition process (Gillooly, 2001). As organisms consume more nutrients due to an increased metabolism, they release more waste products as a result of increased metabolic respiration. It is waste products such as carbon dioxide and methane that contribute further to climate warming.

Freshwater ecosystems are the most impaired of all aquatic ecosystems by anthropogenic factors (Covich et al., 2004), which implies that freshwater systems are the most delicately balanced of all aquatic ecosystems. The experimental freshwater ecosystems exhibited the highest rate of decomposition and the marine the lowest rate at all temperatures, indicating that a saline environment slows the rate of decomposition. Indeed, salt is commonly used in commercial food production to preserve food and prevent spoilage or bacterial colonisation (Ingram & Kitchell, 1967). Previous studies have shown that while low levels of salinity have little to no effect on the decomposition of leaf litter; higher levels do reduce the rate of decomposition when compared to low-salinity environments (Hemminga et al., 1991). High salt levels inhibit the growth of some of the microbial and fungal communities that are essential to the process of decomposition and without them, while the process does continue, it is slowed considerably (Liu & Boone, 1991).

The marine ecosystems at 5 and 10 °C had negative decomposition rates - the mean mass of the leaf matter after seven weeks was greater than the mean mass prior to the study running. Each of the environments that showed a negative decomposition rate were noted to have developed a white fungal growth over the remaining leaf surface. Once the leaves were dried out, the mass of the fungal communities growing on the leaves contributed to the final mass of the leaves, thus giving the appearance of an increase in mass and the resulting negative decomposition rate. While the alder leaves were oven dried prior to their use in the study which in itself sterilised them removing all bacteria and fungal spores, the equipment used in the study was unsterilised, and the temperature controlled rooms were not sterile, allowing for fungal spores or bacteria to enter the environments.

Fungal and bacterial communities play an important role in the decomposition process, “processing” the plant matter for larger macro-detritivores and increasing the nutritional value of the litter (Anderson, 1979). The microbial communities present on decomposing plant matter have been shown to vary with increasing temperature, and so the functional diversity and catabolic profile of microbes present at different temperatures affects not only the rate of decomposition, but also the rate of mineralisation of nutrients in order that they become available to higher trophic levels (Sherman & Steinberger, 2012). Therefore, as temperatures rise it may be that certain fungi or bacteria are removed from the decomposition process entirely, potentially affecting a number of biological processes as well as decomposition, and reducing the carrying capacity (k) or the amount of energy available in the trophic web, inflicting a “bottom-up” effect on the trophic network (O’Connor et al., 2009).

The loss of the Gammarus spp. in many of the experimental ecosystems may have affected the rate of decomposition, providing an overall reduction compared to what might have been had they all survived. However, the macro-detritivores are only one element of the detritus pathway and despite the loss of the amphipods from almost all brackish and marine microcosms, a significant change in mass was still recorded. The loss of the individual gammarids was not recorded throughout the duration of the study and so there is no record of how long each microcosm was subjected to the full level of invertebrate shredding, and due to the lack of control tanks without Gammarus amphipods, it cannot be said how much of the decomposition was due to microbial action and how much could be attributed to the Gammarus. However, recent studies suggest that as global temperatures increase the trophic web for that system will shift to favour those species with a smaller overall body size (Yvon-DuRocher et al., 2010; Forster et al., 2011) and biodiversity will decrease (Covich et al.,
2004). This suggests that as the overall body size of species decreases, then larger species such as *Gammarus* will either begin selecting for smaller individuals or will disappear from the trophic web entirely.

However, as the lost amphipods were only from aquaria containing saline water, it can be deduced that it was the salinity and not the temperature that affected the survival of the *Gammarus*. Those *Gammarus* used in the brackish and marine ecosystems were sourced from a brackish environment, and the marine amphipods were exposed to much higher salinity levels than they normally would be. It may have been that despite efforts to prevent osmotic shock by gradually increasing the salinity levels, the amphipods were still subjected to salinity levels beyond their natural tolerances. As the water used in all freshwater and brackish aquaria was originally sourced from natural water bodies, it may be that a pathogen to which the brackish *Gammarus zaddachi* had no immunity to was spread via the distribution of water from a single source between multiple tanks.

The ratio of alder leaf to stem varied from tank to tank in this set-up, and *Gammarus* amphipods do not consume the stem of the leaves. As such, the quantity of plant matter available to the amphipods varied between microcosms and this may have had a direct influence on the decomposition rate within individual aquaria, although such fluctuations may have had only a very minor effect on the mean decomposition rate of each ecosystem combination. Within the *gammarus* selected to go into the experimental microcosms, there was a variety of sizes and sex ratios which in itself could introduce variation in the rate of leaf matter consumption. Larger individuals may consume greater quantities of plant matter than smaller individuals, altering the overall decomposition rate of each individual aquarium. In addition, the ratio of male to female *Gammarus* may have an affect on the consumption rate of plant matter. For example, three males in a microcosm may consume more plant matter than two males who are spending time ‘displaying’ in order to impress a female in the same system. This study was run without the benefit of a controlled ecosystem without amphipods present. As previously stated, macro-invertebrates are only one element of the decomposition process, and without a control system in place to determine the mean rate of microbial decomposition, the significance of any effects that an increase in temperature has on the metabolism of the *Gammarus* is lost without context.

A number of the microbes involved in the decomposition process are methanogenic bacteria, and if the effects of warming on these methane-producing organisms can be reduced or halted, then that is one side of the warming coin solved as, as previously mentioned, methane is more potent than carbon dioxide as a greenhouse gas.

This paper assesses only one of many side effects of anthropogenic activities, and decomposition is just one of the many ecological processes and biotic factors that will be affected if climate change continues as is currently predicted by the IPCC. With further study of how these environmental factors are affected by global climate change comes a greater understanding of how to reduce the impact of rising temperatures, and if we are to solve the problem on a permanent basis, then it is vital that we understand the cause and effect.
REFERENCES


Silver MW, Gowing MM & Davoll PJ (1986) The association of picoplankton and ultraplankton with pelagic detritus through the water column (0-2000m) Canadian Bulletin of Fisheries and Aquatic Science 214 pp. 311-341


